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EFFECTS OF THERAPEUTIC URINARY CARE DIET TRIAL ON BODY WEIGHT, URINE PROFILE, AND BLOOD PROFILE IN HEALTHY DOGS

By

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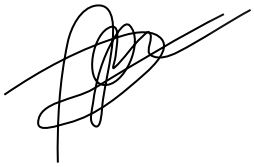

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LIST OF ABBREVIATIONS

RER	Resting Energy Requirement
DER	Daily Energy Requirement
BUN	Blood Urea and Nitrogen
USG	Urine Specific Gravity
UTI	Urinary Tract Infection
LUTD	Lower Urinary Tract Disease
HPF	High Power Field
ME	Metabolized Energy
SPSS	Statistical Product and Service Solutions
TUCD	Therapeutic Urinary Care Diet

ABSTRACT

Urolithiasis is a common urinary tract condition in dogs that can be managed through dietary modification. Therapeutic urinary care diets are formulated to reduce urinary crystal formation and maintain optimal renal health. However, their effects on blood and urine parameters in healthy small-breed dogs remain underexplored. This study aimed to evaluate the short-term effects of a therapeutic urinary care diet (Brand A) on renal biomarkers and urinary characteristics in healthy, male, small-breed dogs. An observational study was conducted in Kelantan, Malaysia, involving five dogs (four small-sized <7 kg and one medium-sized <15 kg). Eligible dogs were male, 8 months to 8 years old, and apparently healthy. The study consisted of three feeding phases: five weeks on a normal diet, one week of acclimatization, and five weeks on the therapeutic urinary care diet. Feed amounts were calculated based on resting and daily energy requirements (RER and DER), with owners instructed to provide two meals daily and ensure constant access to water. Blood and urine samples were collected before and after the trial for paired t-test analysis of renal and urinary parameters, including blood urea and nitrogen (BUN), creatinine, total protein, urine specific gravity (USG), and urine pH. Feeding the urinary care diet for five weeks did not cause any clinically adverse effects. BUN and creatinine levels remained stable ($p > 0.05$), indicating normal renal function. A significant decrease in total protein ($p = 0.038$) and a significant increase in USG ($p = 0.005$) were observed, both within normal physiological limits. Urine pH and crystal presence showed no significant changes. The therapeutic urinary care diet (Brand A) was safe and well-tolerated in healthy small-breed dogs during the short-term trial, maintaining normal renal and urinary parameters. The observed biochemical changes likely reflected expected physiological responses to the diet's composition. Further studies with larger sample sizes and longer durations are recommended to confirm these findings and assess long-term urinary health outcomes.

ABSTRAK

Batu karang atau *urolithiasis* adalah keadaan saluran kencing yang biasa di kalangan anjing yang boleh diuruskan melalui pengubahsuaian diet. Pemakanan penjagaan kencing terapeutik dirumus untuk mengurangkan pembentukan kristal dalam air kencing dan mengekalkan kesihatan buah pinggang secara optimum. Namun, kesannya terhadap parameter darah dan air kencing dalam anjing baka kecil yang sihat masih kurang diterokai. Kajian ini bertujuan untuk menilai kesan jangka pendek pemakanan penjagaan kencing terapeutik (Jenama A) terhadap biomarker buah pinggang dan ciri-ciri kencing dalam anjing jantan baka kecil yang sihat. Satu kajian pemerhatian telah dijalankan di Kelantan, Malaysia, melibatkan lima ekor anjing (empat kecil <7 kg dan satu sederhana <15 kg). Anjing yang layak adalah jantan, berumur 8 bulan hingga 8 tahun, dan sihat. Kajian ini terdiri daripada tiga fasa pemakanan: lima minggu dengan diet normal, satu minggu aklimatisasi, dan lima minggu dengan diet penjagaan kencing terapeutik. Jumlah makanan dikira berdasarkan keperluan tenaga rehat (RER) dan keperluan tenaga harian (DER), dengan pemilik diarahkan untuk memberikan dua hidangan setiap hari dan memastikan air diberikan sepanjang masa. Sampel darah dan kencing diambil sebelum dan selepas ujian untuk analisis Paired-T bagi parameter buah pinggang dan kencing, termasuk nitrogen urea darah (BUN), kreatinin, protein total, ketumpatan urin spesifik (USG), dan pH kencing. Dalam kajian ini, memberi pemakanan penjagaan kencing selama lima minggu tidak menyebabkan sebarang kesan buruk secara klinikal. Tahap BUN dan kreatinin kekal stabil ($p > 0.05$), menunjukkan fungsi buah pinggang yang normal. Terdapat penurunan yang signifikan dalam protein total ($p = 0.038$) dan peningkatan yang signifikan dalam USG ($p = 0.005$), kedua-duanya dalam had fisiologi yang normal. pH kencing dan kewujidan kristal tidak menunjukkan perubahan yang signifikan. Pemakanan penjagaan kencing terapeutik (Jenama A) adalah selamat dan diterima dengan baik dalam anjing kecil yang

sihat semasa ujian jangka pendek ini, mengekalkan parameter buah pinggang dan kencing yang normal. Perubahan biokimia yang diperhatikan mungkin mencerminkan tindak balas fisiologi yang dijangkakan terhadap komposisi diet. Kajian lanjut dengan saiz sampel yang lebih besar dan jangka masa yang lebih lama disyorkan untuk mengesahkan penemuan ini dan menilai hasil kesihatan kencing jangka panjang.

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CHAPTER 1

1.1 INTRODUCTION

The common urinary issue in small animals nowadays is urolithiasis. The word urolith is derived from the Greek word "ouron-" meaning urine, while "-lithos" meaning stone (Tiruneh et al., 2017). According to Foster, 2021, Urolithiasis in the hand simply refers to the formation of uroliths in the urinary system. Canine urolithiasis is not a disease but a clinical sign that is caused by a single or multiple conditions (Tiruneh *et al.,* 2017). The next step after identification of urolithiasis would be determining the underlying systemic disease(s) or any predisposing factors that lead to the condition. There are several types of uroliths in canine, where struvite, calcium oxalate, cysteine, and urate are the most common. According to the analysis of 10 years (2007 - 2016) of epidemiological data of urinary stones in dogs and cats from a referral center in Malaysia by Tajudeen (2017), the prevalence of struvite uroliths in canine is 61.6% with an average age of 2.8 years old. Struvite urolithiasis in dogs is usually caused by urinary tract infection (UTI) initiated by urease-producing bacteria like *Staphylococcus spp*., and *Proteus spp.*. These organisms change the pH in the urinary tract environment from acidic to alkaline which is also known as urine alkalinization and promote the crystallization of magnesium ammonium phosphate. Besides UTI, other predisposing factors such as diet and genetics can also lead to urolithiasis. Formation of calcium oxalate is associated with a diet of high protein, calcium, phosphorus, magnesium, sodium, potassium, and chloride composition (Lekcharoensuk *et al.,* 2002). Although sterile urolithiasis more commonly happens in felines, it can also occur in canines occasionally. In clinical practice, treatment and management for urolithiasis includes medical dissolution, uro-hydropropulsion, surgical removal, less invasive urolith retrieval (catheter retrieval, stone basket retrieval and lithotripsy) and dietary therapy (Foster, 2021). Lately, dietary therapy has risen as an

excellent management and prevention for those dogs with urolithiasis as well as for those with a genetic predisposition. Most of the commercial urinary therapeutic diet helps to maintain a healthy urinary system by controlling the urine pH level at the range of 6.0 - 6.5, limiting the magnesium level, regulating the balance of calcium and phosphorus, maintaining the balance of other minerals, which promotes drinking habits to reduce urine pH concentration, and providing effective antioxidants (Buffington, 2004).

1.2 PROBLEM STATEMENT

The commercialized therapeutic urinary diet for dogs is convenient for the dog patient's owners; however, in a multi-dog household, it is often hard for them to isolate the urolithiasis dog from the other dogs for feeding. Thus, the possible sequelae of a healthy dog consuming a therapeutic urinary diet are a concern among the pet owners as well as the veterinarians. This study seeks to assess the effect of therapeutic urinary care diet on normal healthy dogs through a feed trial. Throughout the study, body weight, body condition score, urine profile, fecal score, and blood profile would be monitored in order to provide empirical evidence on the sequelae of healthy dogs after consuming the therapeutic urinary care diet. The findings from the study would provide a chance for us to understand the effectiveness of dietary therapy as a preventive measure for canine urolithiasis and, at the same time, contribute to a practical perception of possible consequences of routine feeding of therapeutic urinary diet to a normal dog. The outcome aids the feed companies and veterinarians in building proper client education for relevant owners.

1.3 RESEARCH QUESTIONS

1. What is the effect of the therapeutic urinary care diet (TUCD) brand A on the urine profile and blood profile of healthy dogs?
2. What is the effect of TUCD brand A on the body weight and body condition score of healthy dogs?
3. Does providing a therapeutic urinary care diet for urolithiasis predisposed dogs help in preventing the development of urolithiasis?

1.4 RESEARCH HYPOTHEIS

1. There is no effect of TUCD brand A on the urine profile and blood profile of healthy dogs.
2. There is no effect of TUCD brand A on body weight and body condition score of healthy dogs.

1.5 Research Objectives

1. To evaluate the effect of Therapeutic Urinary Care Diet (TUCD) on the urine profile of healthy dogs.
2. To evaluate the effect of TUCD on the blood profile of healthy dogs.

CHAPTER 2

2.0 LITERATURE REVIEW

2.1 OVERVIEW OF CANINE LOWER URINARY DISEASE (LUTD)

Lower urinary tract disease (LUTD) indicates several different conditions that may affect the urinary bladder and prostate gland. The common clinical signs in LUTD dogs are polyuria, polydipsia, hematuria, stranguria, pollakiuria, urinary incontinence or urine burn indicate dysuria that can lead to several possible differential diagnoses which are bacterial urinary tract infection, urolithiasis, micturition disorders related to neurological defects or prostate disease (Mendóza-López *et al.,* 2017). According to the report from Bartges (2004) the most common LUTD in dogs is urinary tract infections (14%) followed by urolithiasis (3%). Bacterial urinary tract infection is detected by clinical manifestations, complete urine analysis, and urine sediment microscopic examination, where results are more than 5 erythrocytes and/or leukocytes per 400x high power field (HPF), together with bacteria and epithelial cells observed suggesting an active inflammation of septic cause (Pressler et al. 2003). Initial antibiotic therapy would be the empirical treatment, but urinary culture would be performed to identify a suitable antibiotic if recurrence bacterial urinary tract infection (Aggarwal & Leslie, 2025). If urolithiasis is suspected, radiography and/or ultrasonography can be proposed as a confirmatory diagnostic test and localized the uroliths which helps in management or treatment planning. Removal of uroliths can be invasive like surgical intervention or minimally invasive by uro-hydropropulsion, intracorporeal laser lithotripsy (Brooks, 2023), basket retrieval and percutaneous cystolithotomy (Foster, 2021).

2.2 ASSOCIATION OF URINARY TRACT INFECTION WITH UROLITHIASIS IN DOGS

Unlike in felines that are usually affected by sterile urolithiasis, canines predispose to “infection stone” which is a urolith formed following urinary tract infection (UTI) (Brister, 2023). The dogs that succumb to UTI are usually immunosuppressed or infected by viral diseases. The etiology of UTI that commonly led to formation of struvite uroliths are urease-producing bacteria mainly *Staphylococcus spp* but also includes *Proteus spp, Pseudomonas spp,* and *Klebsiella spp*. Struvite uroliths are composed of calcium, magnesium, ammonium and phosphate (Waki et al., 2009). The pathophysiology of struvite formation is when the urease produced by the bacteria breaks down the urea in the urine causing the urine pH to increase. As the urea disintegrates, more ammonium and carbon dioxide are released. As the environment in the urinary tract is getting more favorable and presence of building blocks of struvite (ammonium) the struvite crystal accumulates and eventually becomes uroliths (Ivis, 2020).

2.3 OVERVIEW OF UROLITHIASIS AND THE RISK FACTORS IN DOGS

The broad term for the presence of a stone anywhere in the urinary tract is urolithiasis. Urolith in different parts of the body can be named distinctly, for example, kidney stone is nephrolith, bladder stone is cystolith while stone within urethra is ureterolith (Van Vertloo, 2025). Several types of uroliths are present where the most common are calcium oxalate and struvite and less common types which are purine, and cystine stones (Osborne et al., 2009). The risk factors of each type of urolith vary. Formation of calcium oxalate stone is not known but it is suggested that a combination of genetic, dietary and environmental factors lead to hypercalciuria and increase the risk of calcium oxalate precipitation (Houston et al., 2004). Reduction of magnesium, phosphate and citrate in the urine could lead to formation of calcium oxalate formation as they are the inhibitors (Dijcker et

al., 2012). Medical dissolution is not beneficial in calcium oxalate urolithiasis, but dietary therapy plays an important role in the preventive approach (O'Kell et al., 2017). The cause of struvite urolithiasis however is due to urinary tract infection (UTI) as explained above. This type of urolithiasis can be treated using medical dissolution with antibiotic therapy that continues 2 to 4 weeks beyond radiographic or ultrasonographic resolution of uroliths (Weese et al., 2019). Moreover, purine (ammonium urate or xanthine) uroliths are due to metabolic insufficiency which has breed predisposition. For instance, dalmatians, bulldogs and black Russian terriers have a defect in urate transport that leads to hyperuricosuria, thus predisposed to urate urolithiasis (Bannasch et al., 2008). For purine uroliths, medical dissolution is amenable, and it would be more efficient with dietary modification. A diet that helps to alkalinize urine, increase urine volume, and purine-reduced protein (such as eggs and dairy products) can be one of the choices of treatment (Lulich *et al.,* 2016). However, administering allopurinol therapy would have the risk of developing xanthine urolithiasis while increasing urine pH would increase the chance of calcium phosphate urolith formation (Ling et al., 1991). According to Mendóza-López et al., 2017, male (74.22%) dogs are more prone to lower urinary tract disease (LUTD) than female (25.78%) which might be due to the anatomically narrow and long urethra.

2.4 OVERVIEW OF CONTRAINDICATIONS OF URINARY THERAPEUTIC DIET IN DOGS

The awareness among pets' owners about the importance of proper nutrition is now rising in Malaysia causing a shift from homemade meals to commercial pet food products (Vyansaintelligence, 2025). Even some of the owners would prefer premium and specialized dog food products for pet's well-being. The need of a urinary therapeutic diet for the urolithiasis patients is undoubtedly important, but can it be fed to the healthy dogs or fed to the patient in the long term. Although the specialized diet benefits the pet in many ways including weight management, hair and coat support, there are unjustified issues such as the effects on gut-urinary microbiome axis (Salazar et al., 2022). For a very long period, we assumed that urine is sterile in healthy individuals, which is now proven inaccurate as researchers found the presence of urobiome (Li et al., 2017). The potential of gut microflora to contribute in urinary tract diseases can be justified by the ascending urinary tract infection secondary to the perineal contamination by gut microflora (Coffey et al., 2024). The general principle of urinary care diet is promote acidification of urine (pH less than 6.5) which helps in struvite dissolution, reduced in phosphorus and magnesium which are struvite urolith precursors and in some product reduced protein content helps in reducing urea production that result in loss of renal medullary concentration gradient and helps in diuresis (Marcus, 2022). All the functions of the urinary care diet mentioned above will affect the microenvironment of the urinary tract such as urine pH and urinary metabolic parameters, and consequently urobiome communities will be altered (Coffey et al., 2024).

The drawbacks of the urinary care diet was mentioned to be increased risk of other diseases such as congestive heart failure, high blood pressure, and chronic kidney disease due to the high sodium content. (Thornton, 2019). Besides that, the article also suggests that the struvite prevention diets in dogs are only for short term use in diseased, predisposed or recurrence cases. Study shows that increase in salt intake would increase osmotic pressure in the blood vessels causing the intracellular fluid to load extracellularly which increases the mean systemic pressure (Rakova et al., 2017). The cardiac output increases causing the elevation of arterial pressure and induces a baroreceptor reflex that reduces the heart rate. The rising arterial pressure also stretches the small arteries, reducing the total peripheral resistance (Coleman & Guyton, 1969). Besides that, the elevated arterial pressure increases the glomerular filtration pressure, thus increasing the burden on the kidney, leading to higher risk of chronic kidney disease (Acierno et al., 2018). Even an autoregulation mechanism might set in but over time, the hemostasis might not be able to regulate the body system and lead to organ failure (Chandler, 2008). Besides the increased arterial pressure, high sodium intake is also expected to be readily absorbed in the gastrointestinal tract and excreted in the kidney, which proportionally increases kidney workload like glomerular filtration and tubular reabsorption, thus supporting the statement that increased sodium intake increases the risk of chronic kidney diseases (Queau et al., 2020).

Upon diagnosis, switching from conventional diet to urinary diet may be challenging for the owner. The owner may gradually increase the new feed while simultaneously decreasing the old feed (AAHA, 2021). Within this transitional period, the dog requires close monitoring for appetite or dietary indiscretion such as diarrhea, vomiting and allergy reaction (Liao et al., 2023). Moreover, the effect of consuming the urinary care diet by healthy dogs is unknown. This may be

a concern in multidog households, where complete feed separation is challenging to be exercised. Urinary care diets also come at a higher price; thus it may be non-economical especially the multi dog household (Sprinkle, 2025). From the only retail platforms such as Shoppe and Lazada the price of 1 kg urinary care diet is RM 39 to RM 64 which might differ among brand and shop, in comparison to the normal commercial dog kibbles which is around RM 5 to RM 20 averagely.

2.5 OVERVIEW OF AVAILABLE TREATMENT FOR UROLITHIASIS IN DOGS

Uroliths can happen in both upper and lower urinary tract that led to injury of different anatomical counterparts, whereby upper urinary tract consists of kidneys and ureters while the lower urinary tract consists of bladder and urethra (Geddes et al., 2023). The recommended diagnosis and treatments are similar such as medical dissolution, minimally invasive procedures, and invasive procedures but the upper urinary tract involves rapid and invasive intervention and also more advanced diagnosis (Lulich et al., 2016; Osborne & Lulich, 2016). For medical dissolution, it only works on dissolvable uroliths like struvite, cysteine and urate (Osborne et al., 2009). Struvite develops in alkaline urine and urine rich with urease-producing bacteria. Therefore, by neutralizing the urine pH and treating the urinary tract using suitable antibiotics should be able to achieve dissolution (Harris et al., 2023). Cystotomy in struvite urolithiasis should be avoided as there is a high risk of suture-induced urolith recurrence. Urate forms in hyperuricosuria, concentrating urinate and acidic urine, which might be due to failure of purine metabolism. Purine metabolism might be associated with a breed predisposed to defective uric acid transporter and hepatic porto-vascular anomalies (eg, Dalmatians) (Bannasch et al., 2008). If the patient does not have uncorrected liver disease, dissolution is achievable within 4 weeks of a purine-restricted,

alkalinizing, diuretic diet with allopurinol administration. Cystine urolith is due to the inefficiency of proximal tubular reabsorption of cystine thus controlling the protein intake, alkalinizing urine (using potassium citrate or other alkalinizing salt) and 2-mercaptopropionylglycine (Thiola ®) can successfully treat the patient (Rath et al., 2024).

The minimally invasive procedures come in when the urocystoliths accumulate into a big stone and are unable to pass through the urethra. Several newly introduced methods, like intracorporeal laser lithotripsy or percutaneous cystolithotomy, should be opted (Job et al., 2022). Besides the size of the uroliths, some breeds of dog, such as Yorkshire terriers, Maltese and Chihuahua, have relatively small urethras, especially in males, thus minimally invasive procedures can be suggested too (Shigemoto & Kawazu, 2025). Other factors that might affect the selection of procedures would be dissolvability of urolith, experience of the operator, availability of equipment, urolith burden and patient conditions. The rationale of choosing minimally invasive procedures are associated with shorter period of hospitalization and lesser risk of adverse effects compared to invasive surgery (Grant et al., 2008). Combination of medical dissolution with minimally invasive procedures is possible to have a better outcome if the dog is not severely affected. Intracorporeal laser lithotripsy requires an endoscope to penetrate the narrow urethral of the patient which is also known as cystoscope (Wendy, 2023). The cystoscope will release a specific type of laser called a holmium to break the urolith. The fragments of uroliths can be removed via basket retrieval or voiding uro-hydropropulsion (Razvi et al., 1996; (Grant et al., 2008)).

Invasive surgery can be beneficial when a patient requires immediate care either due to life threatening condition or possible deteriorating renal function (Ross et al., 2007). Examples of invasive procedures are subcutaneous urethral bypass in cats, urethral stents in dogs and traditional surgery (such as cystotomy, perineal urethrostomy or prescrotal urethrostomy) (Berent et al., 2014). All of the above procedures aim to achieve instant decompression and halt the side effects like persistent oliguric or anuric, hyperkalemic, progressive azotemia and progressive renal pelvic dilatation (Manassero et al., 2013). Despites, urethral surgeries are not encouraged as the risk of urethral construction is very high. When it is necessary, combination with subsequent extracorporeal shockwave lithotripsy is very helpful to drain the urolith from the urinary tract. In the article written by Lulich et al. (2016), urethral stenting in dog is highly encouraged because it is associated with lower morbidity and mortality rates (<2%), lower reconstruction rate (9%) and also obvious improvement in severity of azotemia in several reports.

2.6 OVERVIEW OF UROLITH PREVENTION

Routine monitoring is one of the best prevention methods, such as urinalysis and a 2 to 3-month interval urine culture as a marker for urinary tract infection that predisposes the animal to urolithiasis (Way et al., 2013). Therapeutic diet as maintenance foods is also recommended due to its low magnesium and phosphorus content which is able to acidify the urine (Queau, 2018). However, patients with calcium oxalate urolith should avoid acidifying the urine as acidic conditions encourage formation of calcium oxalate. In this case, adding thiazide diuretics in the

plan can help to prevent calcium oxalate by enhancing the renal tubular reabsorption of filtered calcium, which reduces urine calcium content (Van Vertloo, 2025).

Some of the commercial therapeutic diet brands claim to have the ability to encourage the patient to drink more, which is possible, but the benefits are short-lived around 3 to 6 months (Queau et al., 2020). After the grace period, the renal function of the patient might be affected due to increasing blood pressure caused by the high dietary salt (Reynolds et al., 2013). Instead, a high moisture diet should be incorporated into the plan to encourage water intake and achieve urine dilution (Stevenson et al., 2003).

Cystinuria is a rare genetic disease that is characterized by renal tubular reabsorption of cystine and commonly prevented by reducing protein intake and alkalinizing the urine (Brons et al., 2013). Another theory of decreasing cystine concentration is by neutering, which is suspected to be an androgen-dependent effect (Giger et al., 2015). This theory is supported by a prospective study, where they observed a significant decrease in urine cystine/creatinine concentration excretion 30 and 90 days after castration (Larsen et al.,2025). Castration is suggested to help in preventing recurrence and genetic transmission of cystine urolithiasis (Brons et al., 2013).

2.7 DEFINITION OF SAFE OR UNSAFE DIET FOR DOGS

Safe diet or food for dogs is defined as nutritionally complete and balanced for specific life stages, free from contaminations of toxic ingredients, and properly prepared and stored. A safe diet should not cause sickness such as vomiting, diarrhea, and discomfort in a dog (Sanderson, 2025). In contrast, an unsafe diet consists of ingredients that bring harm to the dog, such as special toxic food (such as alcohol, chocolate, grapes and tea), heavy metals, mycotoxin or bacteria. Undesired complications like gastrointestinal problems, nervous system disorders, bone marrow suppression and even kidney injuries might arise (Man et al., 2024).

CHAPTER 3

3.0 METHODOLOGY

3.1 STUDY AREA

This study was conducted to dog owners in Kota Bahru, Kelantan, Malaysia.

3.2 STUDY DESIGN

Observational study

3.3 STUDY POPULATION

Dogs in Kelantan, Malaysia which fits the selection criterias.

3.4 SELECTION CRITERIA

3.4.1 INCLUSION CRITERIA

Inclusion criteria are male dogs with age of 8 months to 8 years old and apparently healthy condition. The definition of apparently healthy as stated is dogs manifest vital parameters within normal range and do not exhibit any urinary-related clinical signs such as stranguria, hematuria, dysuria and pollakiuria. As a clarification, the animal that is apparently healthy can be having subclinical issues such as presence of urinary crystals upon microscopic examination of urine.

3.4.2 EXCLUSION CRITERIA

The exclusion criteria for this study are urolithiasis-prone dogs whose owners do not permit their consent, dogs that are clinically unhealthy, and female dogs. Besides that, dogs who had a history of adverse reaction towards foods, dogs that required other therapeutic or special care diets, dogs that were scheduled for surgery during the trial period and dogs that were undergoing other clinical study within the period of time were not recruited.

3.5 SAMPLING METHOD AND PROCEDURE

All enrolled dogs first underwent a physical examination in which body weight, body condition score, and baseline blood and urine profiles were recorded. The feeding trial consisted of three phases. Phase 1 comprised a five-week period during which all dogs were fed their usual diet. This was followed by Phase 2, a one-week acclimatization period during which the dogs were gradually transitioned to the test diet. Phase 3 consisted of the final five weeks of the trial, during which the dogs were exclusively fed the therapeutic urinary care diet (Brand A) at the calculated daily amount. The daily feeding amount had been determined based on each dog's resting energy requirement (RER = 70 × BW^0.75), and the maintenance daily energy requirement (DER) was adjusted according to the predefined activity-factor table.

Table 1: Daily energy requirement (DER) for neutered and intact dogs.

Status	Daily energy requirement
Neutered	DER =1.6 x RER
Intact	DER= 1.8 x RER

The metabolizable energy (ME) content of the therapeutic diet (343 kcal for 100 g) was considered when calculating the feeding amount. Owners were provided with a 50-g measuring cup and were instructed to divide the daily portion into two meals while ensuring that fresh water was available at all times. At the beginning of the trial, owners completed a questionnaire regarding their pet's general health, appetite, behavior, urination, and stool quality. Follow-up assessments were conducted weekly via phone calls to monitor appetite, general condition, urination pattern, and fecal score. Any dog that developed a medical issue during the study period was removed from the trial and referred for appropriate treatment. At the end of Phase 3, all dogs underwent a second physical examination, and blood and urine profiles were collected again to evaluate changes following the dietary intervention. The example of calculation for daily energy requirement is supplemented in Appendix 2.

3.6 Owner Compliance

Owners were requested to ensure that the therapeutic urinary care diet brand A was fed exclusively to the dog participant throughout the entire trial period, particularly in multi-dog households. Owners were also reminded to strictly adhere to the recommended feeding amount without exceeding the calculated daily portion and not to offer any additional food items, including snacks, table scraps, supplements, or other pet foods. Upon any abnormal health conditions observed, the owners may contact the investigator, who will report to the assigned veterinarians. Dogs with conditions requiring intervention that may affect the parameters will be disqualified from the study.

3.7 Data collection tools

During each appointment, the data was collected by using a form and later transferred in the Microsoft Excel. Reference range of urinary parameters and hematological parameters are inserted in Appendix 1.

3.8 Data Analysis

The data is descriptively analysed and tested for normality. Data acquired will be compared with the pre-trial body weight, body condition score, urine and blood profile to determine the effect of therapeutic urinary care diet brand A on each dog. The numerical data will be analyzed by a parametric test and the graph will be plotted using a paired t-test, whereas the categorical data, such as urine pH, urine specific gravity, and presence of crystals in urine will be analyzed using McNemar test to evaluate the hypothesis. Data analysis was conducted using IBM Statistical Product and Service Solutions (SPSS) Statistics (Version 27) . $p<0.05$ is considered statistically significant.

3.9 Ethical considerations

Ethical approvals for using animals (dogs) in the current study was applied for by the Animal Ethics Committee of University Malaysia Kelantan (UMK) (A06.800-1/2/8 JLD 2 (2))

CHAPTER 4

4.0 RESULTS

Pre and post feed trial blood and urine samples are taken from 5 dogs, including 4 small-sized dogs that weigh less than 7 kg and 1 medium size dogs that weigh less than 15 kg. The blood and urine samples were used to evaluate the parameters like BUN, creatinine, total protein, USG, urine pH and formation of crystals.

4.1 Blood and Urine Biochemical Parameters

After five weeks of feeding the TUCD, most biochemical parameters in the dogs showed no significant changes, indicating stable renal and urinary function. Two parameters, however, showed statistically significant differences. BUN has slight decrease from 22.75 to 20.15 mmol/L ($p = 0.237$); creatinine has small increase from 1.74 to 1.81 mg/dL ($p = 0.736$); urine pH has slight increase from 6.50 to 6.75 (p - 0.391) As the significant level is $p < 0.05$, all of them are non-significant changes. Total protein decreased from 6.90 to 6.30 g/dL ($p = 0.038$); USG increased from 1.031 to 1.047 ($p = 0.005$), where both are significant. However, all values remained within the physiological reference ranges. The results are summarized in Table 1.

Table 2: Paired t-test results for pre- and post-feeding biochemical parameters.

Parameter	**Pre-Feeding (Mean ± SD)**	**Post-Feeding (Mean ± SD)**	***t* (df = 3)**	***p* (2-tailed)**
BUN (mmol/L)	22.75 ± 4.50	20.15 ± 1.72	1.474	0.237

Creatinine (mg/dL)	1.74 ± 0.31	1.81 ± 0.38	-0.369	0.736
Total Protein (g/dL)	6.90 ± 0.08	6.30 ± 0.36	3.565	**0.038**
USG	1.031 ± 0.020	1.047 ± 0.019	-7.416	**0.005**
Urine pH	6.50 ± 0.58	6.75 ± 0.50	-1.000	0.391

4.2 Urinary Crystal Formation

McNemar's test was used to compare the presence of urinary crystals before and after the feed trial. The change in urinary crystal occurrence was not statistically significant ($\chi^2 = 1.00$, $p = 0.317$). One dog developed crystals post-intervention, but this difference could be attributed to chance, given the small sample size ($n = 4$).

4.3 Pre-feeding and Post-feeding Body weight

Paired-samples t-test analysis showed no significant difference between pre-feeding and post-feeding body weight (t(3) = 1.669, p = 0.194). Although the mean body weight decreased slightly from 5.90 ± 4.70 kg to 5.69 ± 4.48 kg, the change was not statistically significant, and the 95% confidence interval for the mean difference (−0.197 to 0.632 kg) included zero. The effect size was small (Cohen's d = 0.26), indicating a minimal practical impact. These results suggest that the

five-week urinary diet did not influence overall body weight in the dogs, and weight remained stable throughout the feeding period.

Table 3: Paired t-test results for pre- and post-feeding body weight.

Parameter	**Pre-Feeding (Mean ± SD)**	**Post-Feeding (Mean ± SD)**	**t (df = 3)**	**p (2-tailed)**
Body Weight (kg)	5.90 ± 4.70	5.69 ± 4.48	1.669	0.194

CHAPTER 5

5.0 DISCUSSION

The present study evaluated the short-term effects of a urinary diet on blood and urine parameters in healthy small-breed dogs.

The BUN and creatinine levels remained stable, suggesting that renal function was unaffected by the dietary intervention. The significant decrease in total protein was within normal range; hence it may be associated with dietary composition or increased water intake. A urinary diet has a lower protein composition to help in reducing urinary uric acid and ammonia excretion, which contribute to mixed urolith formation (Stevenson, 2002).

Interestingly, a significant increase in urine specific gravity (USG) was observed. This may reflect changes in urine concentration due to the nutritional formulation or altered hydration status. Such changes are expected in specialized urinary diets that modulate mineral solubility to support urinary tract health. Urinary diet will be formulated with higher dietary sodium, which stimulates thirst by the complex hemostasis mechanism, such as the release of antidiuretic hormone (ADH), and results in higher urine output and more diluted urine (Queau *et al.,* 2020).

The urine pH showed a non-significant mild increase, while urinary crystal presence did not change significantly after the feeding period. The changes in urine pH were not significant because they are within the normal range of 6 to 7.5 (Rizzi, 2014). One dog developed crystals post-trial; however, this may be attributable to other confounding factors such as water intake and feed adjunct to the main diet, i.e., snacks. A larger sample size is required to further elucidate these

findings. Body weight also remained stable throughout the study period, with no significant difference between pre- and post-feeding values.

Overall, these findings indicate that the urinary diet was safe and well-tolerated in healthy dogs, with no evidence of renal or metabolic side effects during the short-term feeding period. However, the study's small sample size (*n=4*) limits the statistical power, and further studies with larger sample populations and longer duration are recommended to confirm these observations.

CHAPTER 6

6.0 CONCLUSION AND RECOMMENDATION

This study demonstrated that short-term feeding of a urinary diet for five weeks in healthy small-breed dogs did not produce any adverse effects on renal or urinary parameters. Blood urea nitrogen (BUN) and creatinine levels remained stable, indicating normal kidney function, while the observed reduction in total protein and increase in urine specific gravity reflected expected physiological responses to the dietary formulation rather than pathological changes. Urine pH and crystal presence showed no significant alterations, suggesting that the diet effectively maintained urinary health without predisposing to crystalluria. Overall, the urinary diet was safe and well-tolerated under the study conditions.

The findings also suggest that, in multi-dog households, the therapeutic urinary care diet could be safely provided to all dogs, even if only one dog requires urinary support. This approach offers practical benefits, including increased convenience for the owner, reduced time and effort in meal management, and enhanced animal welfare, as dogs are social animals that thrive on interaction with their group. Moreover, these results provide veterinarians with confidence in recommending such diets without inadvertently causing health issues in otherwise healthy dogs.

However, given the small sample size and limited trial duration, further research with a larger population and extended feeding period is recommended to confirm these findings, evaluate potential long-term effects, and further support dietary recommendations for both clinical and multi-dog household settings.

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8.0 APPENDIX

Appendix 1

Reference range of urinary parameters in dogs.

Urinalysis Parameters	Normal Value
Chemical determination	
pH	4.5-8.5
Specific Gravity	1.001 - 1.065
Glucose	Negative
Ketone	Negative
Bilirubin	Trace to 1+
Protein	Trace
Urobilinogen	Trace
Haemoglobin	Negative
Microscopic determination	
RBC (cells/HPF)	0-5
WBC (cells/HPF)	0-5
Casts (casts/LPF)	Occasional Hyaline
Epithelial cells (cells/HPF)	Occasional
Bacteria	Negative
Crystals eg: Calcium oxalate monohydrate Calcium oxalate dihydrate Calcium carbonate	variable
Others Spermatozoa Microfilariae	Present or absent

HPF high power field, LPF low power field

[a]Osborne et al. (1995)

Reference range of hemogram in dogs.

Haemogram	Ref.
WBC (10^3/µl)	6.0-15.5
LYM	1.0-5.0
MON	0.1-1.0
GRA (10^3/µl)	2.0-8.0
LYM%	20.0-55.0
MON%	2.0-12.0
GRA%	35.0-75.0
RBC (10^6/µl)	6.5-10.00
HGB (g/dl)	10.0-15.0
HCT %	30.0-45.0
MCV (µm^3)	39.0-55.0
MCH (pg)	25.9-34.0
MCHC (g/dl)	30.0-35.9
RDW (%)	10.0-16.0
PLT (10^3/ µl)	300-800
MPV (µm^3)	7.0-11.0
PCT (%)	0.200-0.500
PDW (%)	10.0-18.0

Reference range of hemogram in dogs.

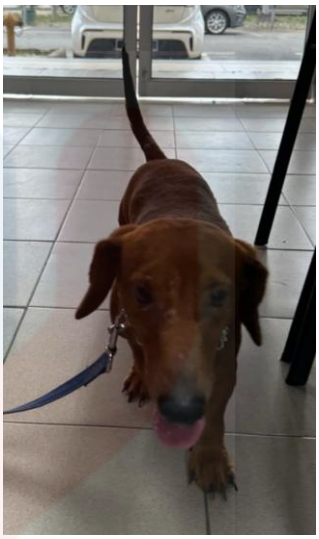
Biochemistry		Ref.
ALB	g/dL	2.2 – 4.0
ALKP	U/L	14 – 111
ALT	**U/L**	12 – 130
BUN	mg/dL	16 – 36
CREA	mg/dL	0.8 – 2.4
TP	g/dL	5.7 – 8.9
TRIG	mg/dL	10 – 100
GLOB	g/dL	2.8 – 5.1

Appendix 2

Example
10kg neutered dog RER = 70 x 10 $^{0.75}$ = 393.6 kcal/day **DER (Neutered) = 1.6 x RER = 629.76 kcal/day** Feed amount during weight loss = 629.76 kcal/day ÷ 343 kcal/100g = 183.6 g/day Number of 50g cups of feed given per day = 183g/50g = 3.66 cups/ day ~ 3 cups/ day Number of 50g cups of feed to be given per meal = 3 cups/ 2 meal = 1.5 cups per meal, twice daily **DER (Intact) = 1.8 x RER = 708.48 kcal/day** Feed amount during maintenance = 708.48 kcal/day ÷ 343 kcal/100g = 206.5 g/day Number of 50g cups of feed given per day = 206g/ 50g = 4.12 cups /day ~ 4 cups / day Number of 50g cups of feed to be given per meal = 4 cups/ 2 meal = 2 cups per meal, twice daily

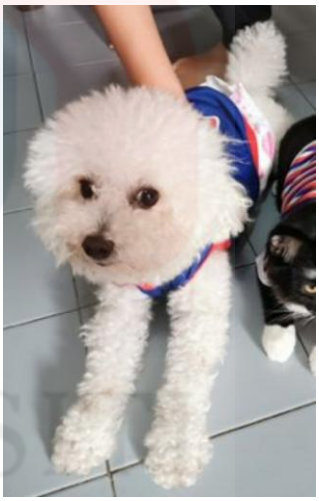
Appendix 3

Picture 1: LiLi

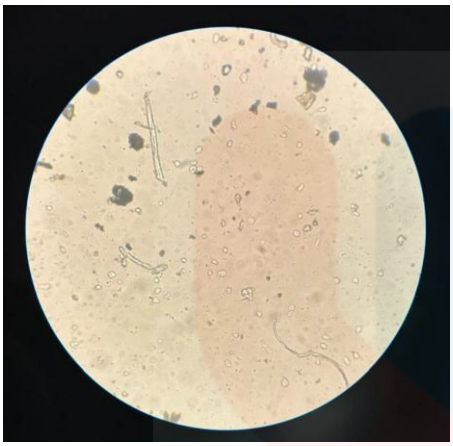


Picture 2: Happy

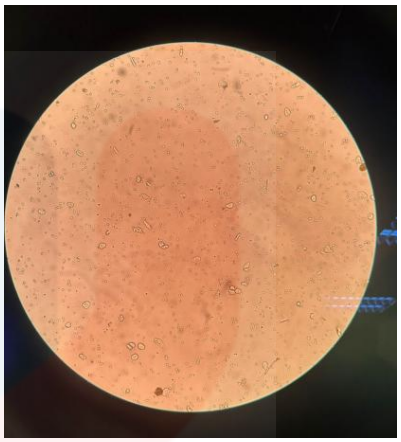
Picture 3: Jack



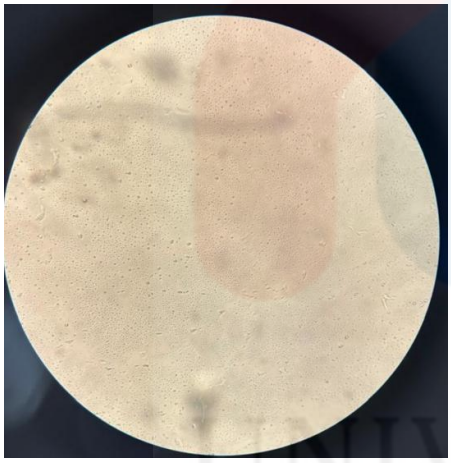
Picture 4: Snoopy



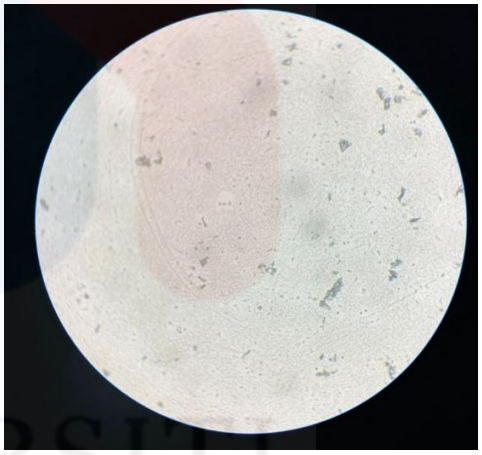
Picture 5: Lili's pre feed trial urine cytology



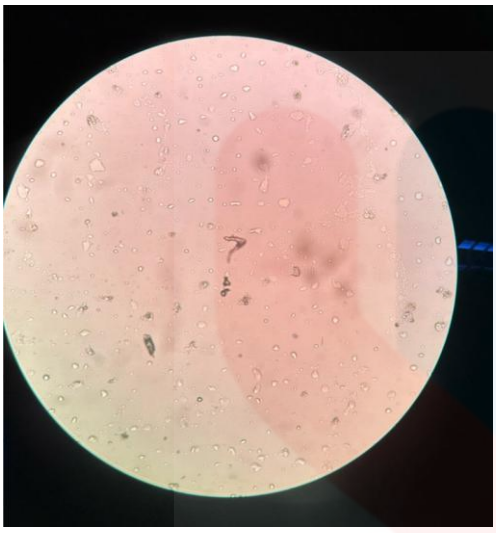
Picture 6: Lili's post feed trial urine cytology



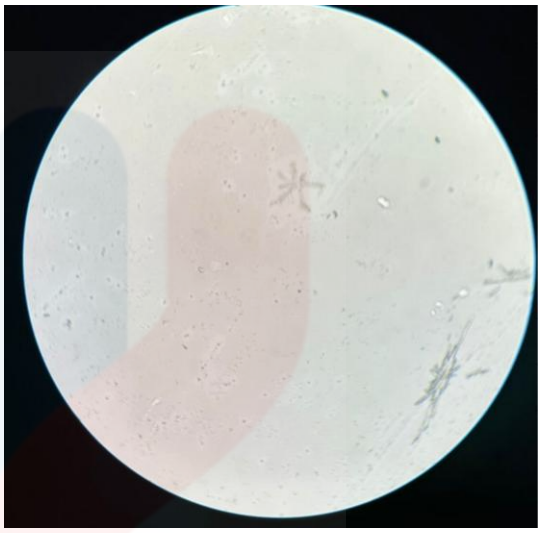
Picture 7: Happy's pre feed trial urine cytology



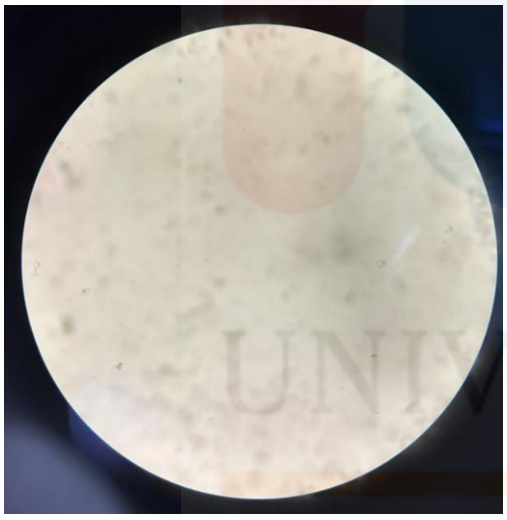
Picture 8: Happy's post feed trial urine cytology



Picture 9: Jack's pre feed trial urine cytology



Picture 10: Jack's post feed trial cytology



Picture 11: Snoopy's pre feed trial urine cytology

Picture 12: Snoopy's post feed trial cytology